



IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re application of:  
Masaharu NODA et al.

Appl. No. 09/920,653

Confirmation No. 6043

Filed: August 3, 2001

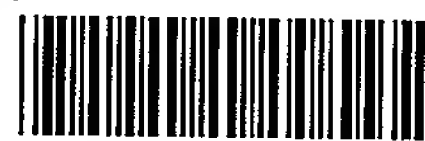
For: NAV2 CHANNEL GENE-DEFICIENT  
NON-HUMAN ANIMALS

Art Unit: 1362

Examiner: Not Yet Assigned

Atty. Docket No. 31671-173164

Customer No.



26694

PATENT TRADEMARK OFFICE

**Second Preliminary Amendment**

Assistant Commissioner for Patents  
Washington, D.C. 20231

Sir:

Prior to calculation of the fees, please amend the specification as follows:

**IN THE SPECIFICATION**

On page 35, replace the paragraph beginning at line 10 with the following:

-- Next, nerve cells in dorsal root ganglia were isolated. The dorsal root ganglia were prepared from wild-type and  $Na_v2$  gene-deficient mice of 8-16 weeks of age. Nerve cells were dispersedly isolated from the dorsal root ganglia according to the method of Renganathan et al. (J Neurophysiol 84, 710-718, 2000). Before used for an ion imaging experiment, the dispersedly isolated nerve cells were cultured under the condition of the humidity of 100% and the temperature of 37°C, and with 5% of carbon dioxide, then adhered to the glass of culture plates. All nerve cells were confirmed to be  $Na_v2$ -positive by staining

nerve cells of dorsal root ganglia derived from wild-type mice with the above-mentioned anti- $\text{Na}_v2$  antibody. The size of the dispersedly isolated nerve cells were comprised of 3 groups of small (25 micron or smaller in diameter: about 50%), medium (25 to 40 micron in diameter: about 40%), and large (40 micron or larger in diameter: about 10%). However, there was no difference between the materials isolated from wild-type and gene-deficient mice in the size, shape and survival rate of these 3 types of cell. The survival rate was verified by Tripan blue staining. –

On page 39, after the first full paragraph delete the remainder of the page

Delete pages 40 and 41 in its entirety.

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**REMARKS**

This Preliminary Amendment is made to correct a typographical error and to eliminate the unnecessary sequence listing on pages 39 – 41. Examination on the merits of the application is requested. A marked up version showing the changes made is attached.

Date:

10/16/01

Respectfully submitted,



Robert Kinberg

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10/16/01 10:00 AM



Applicant(s) Masaharu NODA et al.  
Appl. No. 09/920,653

**VERSION WITH MARKINGS TO SHOW CHANGES MADE**

Next, nerve cells in dorsal root ganglia were isolated. The dorsal root ganglia were prepared from wild-type and  $Na_v2$  gene-deficient mice of 8-16 weeks of age. Nerve cells were dispersedly isolated from the dorsal root ganglia according to the method of Renganathan et al. (J Neurophysiol 84, 710-718, 2000). Before used for an ion imaging experiment, the dispersedly isolated nerve cells were cultured under the condition of the humidity of 100% and the temperature of 37°C, and with 5% of carbon dioxide, then adhered to the glass of culture plates. All nerve cells were confirmed to be  $Na_v2$ -positive by staining nerve cells of dorsal root ganglia derived from wild-type mice with the above-mentioned anti- $Na_v2$  antibody. The size of the dispersedly isolated nerve cells were comprised of 3 groups of small (25 micron or smaller in diameter: about 50%), medium (25 to 40 micron in diameter: about 40%), and large (40 micron or larger in diameter: about 10%). However, there was no difference between the materials isolated from wild-type and gene-deficient mice in the size, shape and survival rate of these 3 types of cell. The survival rate was verified by Tripan blue staining.

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